

## WHAT IS CLAIMED IS:

Sub A 5  
1. Imaging apparatus comprising an optical assembly, a light source assembly and an imaging detector, said assemblies being operative to provide an image at a TIR surface defined by said optical assembly, said light source assembly directing polarized light at said TIR surface in a manner to generate an evanescent field at said surface, said optical assembly having spaced-apart, top and bottom surfaces, said bottom surface including first and second gratings, said gratings being located and configured to direct light  
10 passing through the first grating from said source to said top surface and light reflected from said top surface passing through to said second grating to said detector respectively to cause TIR and an evanescent field associated therewith by a single reflection at the TIR surface.

15 2. Apparatus as in claim 1 wherein said first and second gratings comprise Bragg gratings.

3. Apparatus as in claim 1 wherein said gratings comprise electrically alterable gratings, said apparatus also including means for altering said gratings in a manner to change the direction of light incident thereto.

20 4. Apparatus as in claim 3 wherein said gratings comprise electrically alterable holograms.

5. Apparatus as in claim 1 also including a wavelength filter for selecting the wavelength of light from said light source.

25 6. Apparatus as in claim 1 also including an arcuate adjusting mechanism for moving said light source about an arcuate path to selected positions therealong to alter the angle of the light into the optical element.

7. Apparatus as in claim 1 including a flow cell secured to said TIR surface, said flow cell including an inlet port and an outlet port for flowing analyte across said TIR surface.

30 8. Apparatus as in claim 1 also including a computer, said computer including means for converting images from said reflected light into a convenient form for analysis.

9. Apparatus as in claim 5 wherein said filter means comprises a filter wheel located to intercept light from said light source, said apparatus including means for rotating said wheel.

10. Apparatus as in claim 5 wherein said filter means comprises a stack filter.

5 11. Apparatus as in claim 3 also including a plurality of imagers, each of said imagers being located at a position to capture an image generated in response to input light at a corresponding angle.

10 12. Apparatus as in claim 5 also including a plurality of imagers each of said imagers being located at a position to capture an image generated in response to input light of a corresponding wavelength.

13. Apparatus as in claim 7 wherein said flow cell includes means for securing a slide thereto.

15 14. A cassette for use with a TIR analyzing instrument, said instrument comprising a processing assembly having a polarized light source portion providing a polarized light beam, a reflected light analyzing portion and a locating portion for locating the cassette for TIR imaging of a specimen array on an optical element of said cassette comprising;

20 an optical element comprising an upper TIR surface on which a specimen array is formed, said optical element being configured to receive polarized light directed to the TIR surface and generating an evanescent field at the TIR surface;

a mount or frame coupled to the optical element and having a locating portion engageable with a mating locating element of the TIR instrument said mount or frame being operable to locate at least a selected portion of the specimen array in the evanescent field.

25 15. Imaging apparatus as in claim 14 wherein said TIR surface comprises the upper surface of a transparent slide said slide having a lower surface, said lower surface comprising at least one grating and said upper surface including an array of receptors thereon.

30 16. Apparatus as in claim 14 wherein said at least one grating comprises a first grating positioned to intercept said polarized light beam from said light source portion

and a second grating positioned to intercept said light beam after it has reflected from said upper TIR surface.

17. Apparatus as in claim 14 wherein said cassette and said processing assembly are configured for a single reflection of the light beam at the TIR surface.

5 18. Apparatus as in claim 14 further including a flow cell secured to said TIR surface, said flow cell including an inlet port and an outlet port for flowing analyte across said specimen array.

19. Apparatus as in claim 15 wherein at least one grating comprises an electrically alterable grating.

10 20. An imaging apparatus for imaging a specimen array within the evanescent field present upon reflection of a beam of light at a TIR surface comprising;

15 a polarized light source emitting a polarized extended beam of light;  
an optical element having a TIR surface on which the specimen array is placed and a surface having grating portions, the optical element being placed to cause the beam of light to pass through a first grating portion to direct the light to the TIR surface for being reflected only a single time at the TIR surface, the specimen array being within the evanescent field associated with the total internal reflection at the TIR surface and the beam of light after said single reflection passing through a second grating portion, said gratings being configured to pass the beam of light through them to the TIR surface and from the TIR surface to enable TIR effect to occur; and

20 a polarization – sensitive, imaging detector, said detector detecting the beam of light reflected from the TIR surface including the spatially distributed polarization change caused by the specimen array.

25 21. The imaging apparatus of claim 20 wherein the light source and imaging detector are constructed as a first separate assembly defining a processing assembly and the optical element is part of a second separate assembly defining a cassette and the cassette is removably fitted to the processing assembly.

30 22. The imaging apparatus of claim 21 wherein the cassette comprises the optical element having an upper surface defining the TIR surface on which the specimen array is

placed and a lower surface having the first grating portion and the second grating portion each grating portion positioned so that the beam of light from the processing assembly will pass through the first grating portion into the optical element and after reflection will pass through the second grating portion prior to exiting the optical element.

5        23. The imaging apparatus of claim 22 wherein the cassette has a mount or frame portion to which the optical assembly is attached and the mount or frame portion has mating portions and the processing unit has mating/receiving portions such that by matingly fitting the mating portions to the mating receiving portions the cassette is removably fitted to the processing assembly.

10        24. The apparatus of claim 23 in which at least one of said grating portions is electronically alterable and further comprising controllable electronics altering means for electronically controlling at least one grating whereby the angle of the beam of light exiting from the at least one electrically alterable grating may be varied.

15        25. The apparatus of claim 23 in which the light source in the processing assembly may be changed by an arcuate adjusting mechanism to be directed at selected different angles toward the optical element of the specimen assembly to alter the angle of the light into the optical element.

20        26. The apparatus of claim 23 wherein said and said processing assembly are movable in relation to each other to enable selecting portions of the specimen array to be sequentially imaged.

25        27. Apparatus of the type in which total internal reflection of a light beam occurs at a TIR surface in which an evanescent field is created, by passing a beam of light into an optical member or assembly of members defining an optical portion the improvement comprising;

a first grating at a surface of the optical portion at which the light beam is directed into the optical portion the first grating being configured to direct the light beam through the optical portion to the TIR surface to cause total internal reflection at the TIR surface.

a second grating at a surface of the optical portion the second grating being configured for the light beam to exit the optical portion through the second grating.

28. The apparatus of claim 27 in which the apparatus has material under investigation in the evanescent field and said material causes a change in polarization of the light beam.

29. The apparatus of claim 27 in which the light beam exits the optical portion  
5 after a single reflection at the TIR surface.

30. The apparatus of claim 27 in which the light beam is polarized prior to entering the optical portion.

31. The apparatus of claim 30 in which the polarized light beam has a first coherent length and the optical portion has a first distance between the first grating and  
10 the TIR surface and second distance between the TIR surface and the second grating, said first and second distances each being smaller than said first coherent length.

32. Imaging apparatus comprising an optical assembly, a light source assembly and an imaging detector, said assemblies being operative to provide an image at  
15 a TIR surface defined by a top surface of said optical assembly, said light source assembly directing at said TIR surface polarized light having a first coherent length in a manner to generate an evanescent field at said TIR surface, said optical assembly having a bottom surface spaced apart from said top surface a distance smaller than said first coherent length said bottom surface including first and second gratings, said gratings  
20 being located and configured to direct light passing through the first grating from said source to said TIR surface and light reflected from said TIR surface passing through to said second grating to said detector respectively to cause TIR and an evanescent field associated therewith by a single reflection at the TIR surface.

33. Apparatus as in claim 32 wherein said first and second gratings comprise Bragg gratings.

25 34. Apparatus as in claim 32 wherein said gratings comprise electrically alterable gratings, said apparatus also including means for altering said gratings in a manner to change the direction of light incident thereto.

35. Apparatus as in claim 34 wherein said gratings comprise electrically alterable holograms.

30 36. Apparatus as in claim 32 also including a wavelength filter for selecting the wavelength of light from said light source.

37. Apparatus as in claim 32 also including an arcuate adjusting mechanism for moving said light source about an arcuate path to selected positions therealong to alter the angle of the light into the optical element.

38. Apparatus as in claim 32 including a flow cell secured to said TIR surface, said flow cell including an inlet port and an outlet port for flowing analyte across said TIR surface.

39. Apparatus as in claim 32 also including a computer, said computer including means for converting images from said reflected light into a convenient form for analysis.

40. Apparatus as in claim 36 wherein said filter means comprises a filter wheel located to intercept light from said light source, said apparatus including means for rotating said wheel.

41. Apparatus as in claim 36 wherein said filter means comprises a stack filter.

42. Apparatus as in claim 32 also including a plurality of imagers, each of said imagers being located at a position to capture an image generated in response to input light at a corresponding angle.

43. Apparatus as in claim 36 also including a plurality of imagers each of said imagers being located at a position to capture an image generated in response to input light of a corresponding wavelength.

44. Apparatus as in claim 38 wherein said flow cell includes means for securing a slide thereto.

45. Imaging Apparatus comprising an optical assembly, a light source assembly and at least one imaging detector, said assemblies being operative to provide an image reflected from a TIR surface defined by a top surface of said optical assembly, said light source assembly being operative to direct at said TIR surface polarized light in a manner to generate an evanescent field at said TIR surface, said optical assembly having a bottom surface, said bottom surface including first and second spaced apart gratings, said grating being located and configured to direct light passing through said first grating from said source to said TIR surface and light reflected from said TIR surface to said second grating to said detector respectively, said optical assembly being configured to

impose on said light reflected from said TIR surface a superposition of images therefrom to provide a single reflection therefrom.

46. Apparatus as in claim 45 wherein said light is monochromatic.

47. Apparatus as in claim 45 including filter means for selecting a single

5 image exiting said second grating.